A VARIATION OF GEMMATION OF BLASTOMYCES DERMATITIDIS IN THE TISSUE LESION *

PLATES 22 AND 23

H. WINDSOR WADE

From the Pathological Department of Charity Hospital, and the Department of Pathology and Bacteriology of Tulane University, New Orleans, Louisiana

While a series of cases of systemic blastomycosis was being studied microscopically, there was encountered in a skin lesion of one of these a condition so unusual and at first so difficult of explanation that it was made the subject of a particularly careful morphologic study. Tho there were numerous budding organisms typical of the so-called blastomyces, the presence of numerous minute forms at first suggested sporulation. This peculiarity may be emphasized by a brief description of the typical appearances of the parasites found in the lesions of the recognized yeast-mold infections.

Tho originally recovered from the more common skin affection and named accordingly, Blastomyces dermatitidis (Gilchrist and Stokes[1]) is the cause of systemic blastomycosis. It appears in the tissues as spherical, ovoid, or irregular bodies possessing doubly contoured highly refractile cell membranes, which vary considerably in thickness in different individuals. The parasites show considerable variation in size, the average being from 12 to 20 microns in diameter. The protoplasm is irregularly granular, at times vacuolated, and is as a rule acidophilic, tho the contained granules often stain with the basic dyes. These granules are apparently not connected with spore-formation, and this parasite is held to multiply in the tissues only by budding. During the process of formation and subsequently the buds are seldom less than from 4 to 6 microns in diameter, and can be clearly made out with the ordinary dry lenses. Coccidioides immitis (Rixford and Gilchrist[2]), the causative agent of coccidioidal granuloma, has been shown to be a distinct type of fungus, most recently by McNeal and Taylor[3] and by Brown and Cummings.[4] Altho in the tissue lesions it resembles the blastomyces somewhat, it usually appears larger and less hard-walled. The individuals vary more widely in size, ranging from 5 to 50 microns in diameter. The protoplasm may be finely granular or vacuolated, or may be segmented into numerous irregular daughter cells. Other large bodies are filled with numerous smaller spheres or spores, usually flattened where apposed, and each surrounded by its individual capsule. Large ruptured capsules are commonly found in the lesions but it is said that the small

* Received for publication December 1, 1915.

1 Jour. Exper. Med., 1891, 3, p. 53.
2 Bull. Johns Hopkins Hosp., 1896, 1, p. 209.
3 Jour. Med. Research, 1914, 30, p. 261.
4 Arch. Int. Med., 1915, 15, p. 608.

parasites, either naked or encapsulated, are not recognizable ordinarily until they have enlarged somewhat and the distinguishing capsule has developed. Multiplication by budding is said never to occur in this organism, and it is now generally accepted as a species distinct from the blastomyces.

In but few reports in recent literature was it thought that indications of both budding and sporulation of the parasites had occurred in the tissue lesions or exudates. LeCount and Myers[5] studied microscopically and carefully described the lesions in a case of systemic blastomycosis originally reported by Eisendrath and Ormsby.[6] Widespread lesions were found, in all but one of which the organisms were of the ordinary type. In the cerebellum, however, was an area in which were parasites thought to have multiplied by endosporulation. Very minute, blastomycetes occurred in dense clusters numbering several hundreds. Among these there were a number of budding cells, but not enough to account for the great numbers of minute forms. In some of the aggregations large torn capsules were found, but it was said that intermediate forms of development between the supposed spores and the mature organisms were not demonstrated.

Montgomery[7] described a case which, while not identical with that recounted, is at least similar in that the organism present was thought to multiply both by budding and by spore-formation. In nearly every tube inoculated from unbroken abscesses pure cultures of the blastomyces were obtained, and the organism was unusually pathogenic for guinea-pigs. Nevertheless, in the smears and sections from both man and animal, there were found exceedingly few organisms in the ordinary forms. There were, on the other hand, large numbers and masses of round cells, each about the size of a red blood corpuscle, strongly resembling small blastomycetes but without the usual double-contoured capsule. No large sporulating forms could be found. Montgomery suggested in this publication that these were possibly young rapidly multiplying organisms, tho no relation could be traced between the small bodies and the larger budding forms occasionally met with. In his publication with Ormsby,[8] however, this case was summarized and the statement made that these organisms were thought possibly to have been produced by a process of spore-formation.

In several other instances unusual variations in the morphology of the blastomyces are described, but not completely enough positively to establish distinct type variations in the infecting organisms. Hektoen[9] described very minute forms which developed into the larger. Ricketts[10] mentions finding in a teased specimen of excised tissue a small form, found together with the typical organisms. These smaller bodies occurred in pairs and chains, were encapsulated, and measured about 3.75 microns in diameter. Their nature and significance were not discussed. Another inconclusive, but very interesting, description was made by A. J. Smith[11] of a small type of organism which by special methods stained differentially both from the ordinary blastomyces and from body cells. They were round to oval, with thick, single, deeply staining basophilic capsules and lightly basophilic centers, the latter containing several vacuoles suggesting endogenous spores of yeasts. Because of lack of fresh material, these organisms could not be studied culturally. In

[5] Jour. Infect. Dis., 1907, 4, p. 187. Tr. Chicago Path. Soc., 1907-8, 7, p. 49.
[6] Jour. Am. Med. Assn., 1905, 45, p. 1045.
[7] Jour. Cutan. Dis., 1907, 25, p. 393.
[8] Arch. Int. Med., 1908, 2, p. 1.
[9] Jour. Exper. Med., 1899, 4, p. 261.
[10] Quoted by Montgomery, Jour. Cutan. Dis., 1901, 19, p. 38.
[11] Med. Bull. Univ. of Pennsylvania, 1909-10, 22, p. 362.

a case reported by Hildreth and Sutton[12] from Porto Rico, there were observed beside numerous typical cells, many of which were budding, numerous smaller spherical bodies about 3 microns in diameter. No further description of these is made.

In the skin lesions of a case which will be reported more fully elsewhere has been found a condition more particularly like that described by LeCount and Myer, in which there were vast numbers of minute parasites very suggestive of spores. Study of this lesion in specially stained preparations demonstrates a condition of the organisms present which has not, so far as can be learned, heretofore been accurately described.

The essential features of the case may be summarized as follows: A colored man, 18 years of age, presented himself at the hospital with a dermatitis, thought to be pellagrous, over hands, wrists, neck, and ankles. In subcutaneous tissues of the inguinal region there was an added infiltrative lesion of tubercular type. Over this the epithelium was thickened and rough and showed numerous small fissures covered by encrusted exudate. In places a yellowish purulent material could be expressed from the deeper tissues. Tubercle bacilli were found in the sputum. At autopsy (Dr. T. D. Hurley) lesions typical of tuberculosis were found in the lungs. Sections of the lung tissue showed extensive granulomatous lesions with necrosis, in many of which tubercle bacilli were demonstrated. Here and there, however, a number of typical encapsulated blastomycetes occurred. A few were lying free but the majority were intracellular. All were mature thick-walled forms, very few of which showed evidence of budding. No other deep organ was found to be involved in either the tuberculous or the blastomycotic process.

Skin Lesion.—In sections of skin taken from the inguinal region there was a condition which superficially resembled the lesion of coccidioidal granuloma as well as that of blastomycosis. The epidermis showed little of the usual downward proliferation and the parasites were for the most part in the deeper layers. In the firm connective tissue were lesions which appeared to be abscesses, immediately about which the chorium was partially degenerated and more or less infiltrated with lymphoid and plasma cells. Few polymorphonuclear leukocytes were found. Within such an area and merging into the central region was a zone which was of unusual type. This area at first glance was suggestive of ordinary necrosis, was seen to be made up very largely of rather unusual "endothelioid" cells (endothelial leukocytes) which had for the most part fused to form great numbers of giant cells (Fig. 1). These giant cells presented an unusual appearance; they were often very large and presented coarsely granular appearance (Fig. 2). So numerous were they that the central broken down material seemed often to have been derived entirely from their disintegration. This process was not entirely an infiltration by endothelial leukocytes, but was also degenerative. This was shown not only by the general appearance of the affected skin, but by the fact that in the lesions well-formed blood-vascular spaces of fair size were seen at times quite unsupported by other than the described cells of the granuloma (Fig. 1). These vessels

[12] Jour. Am. Med. Assn., 1914, 63, p. 2289.

were not at all the new vessels of granulation tissue. A feature of the lesion was that, notwithstanding the amount of endothelial-cell infiltration, giant-cell-formation, and necrosis together with, as will be described, the presence of vast numbers of blastomycetes, there was often practically no fixed-tissue reaction or invasion by leukocytes of the ordinary types. Where such infiltration was present, it was not commensurate in extent with the lesion.

Scattered throughout this lesion, among the cells and cell debris but particularly within the giant cells, were great numbers of blastomycetes of all sizes and of varying appearance (Figs. 3, 4, and 5). Numbers of these were in the common spherical and budding forms of the organism. Beside these, however, there were seen, on close scrutiny, vast numbers of organisms so small and so massed as to give to the giant cells and necrotic material their coarsely granular appearance. This is seen particularly well in Fig. 2. Very many of these minute organisms measured about 1.5 to 3 or 4 microns in diameter and frequently could be defined only as large granules, many of which were indistinguishable from the protoplasmic granules of the surrounding structures. Only the larger of these small organisms could clearly be seen, in ordinarily stained specimens, to possess capsular membranes. In the outer zones of a focus of infection this minute form of the organism was usually absent, or present in small numbers. Nowhere were tubercle bacilli demonstrable in specially stained sections.

Altho the parasites differed widely in appearance from those found in the tissues of the coccidioidal granuloma, it was thought that the small forms were produced by some process of spore-formation, and careful search was made for ascospores. Occasional broken shell-like structures similar to those described by LeCount and Myer[5] could be found, some of which contained more or less numerous minute parasites, but no organism was encountered which showed any evidence of endosporulation.

An occasional mature cell was found to be divided into two distinct individuals within the sclerotic outer shell by a transverse wall of the more delicate inner or soft membranous capsule. These daughter cells did not resemble spores, and in no instance was this process seen to further subdivide the organism. From the finding in the special preparations later to be described of two or three organisms which showed incomplete stages of this subdivision, it was concluded that these forms resulted from an attempt at ordinary budding which was restrained by the inability of the primary pseudopodium of the bud to rupture the sclerotic shell. This process may be described as originating by a herniation of the inner capsule at one point, the resulting "pseudopodium" being forced to occupy a position between the mother cell and the hard capsule. This process seems not to have been mentioned in previous descriptions of the organism.

COMPARISON OF STAINING METHODS

In order to determine the best staining technic for the demonstration of the minute organisms in this lesion, as well as for routine use in the study of blastomycotic lesions, a great many preparations were made, and tissues from several cases subjected to many special processes. This work was made necessary by the fact that few of the recent authors on blastomycosis discuss such special stains at any length, and the processes referred to in the literature available at the time, proved inadequate for the purpose. The results obtained will be indicated briefly.

Tho recommended by some, the modifications of Romanowski's stain were employed with but indifferent success. The best of these was Giemsa's. Altho it has been said to stain the capsule a faint blue, in my hands it left both Zenker's and formalin-fixed tissues uncolored. The cytoplasm usually assumes a blue or purplish color which is but slightly different from that of the cell nuclei. Wright's and Leishman's stains were less satisfactory. Even the eosin and methylene-blue technic of Mallory, tho it sometimes gives very pretty results and is said by Wolbach[13] to be the best for demonstrating the coccidioides in tissues, failed to give clearly differential results in the special lesion studied. Unna's polychrome methylene blue was similarly disappointing.

In view of the usefulness of allied processes in the study of smear preparations of certain yeast organisms, as described by Verity[14] and others, it had been hoped that Levaditi's silver-impregnation method, or some modification of this, would prove differential for the capsules of the blastomycetes. In the tissues available, however, which had been in formalin for some time, this method was also without value. It was also expected that at least certain forms of the blastomycetes would exhibit affinity for stains used in demonstrating amebae in the tissues. This proved not to be the case. It may here be added that the results with Van Gieson's stain and with Mallory's phosphotungstic-acid hematoxylin stain were similarly unsatisfactory, tho the protoplasmic granules were made very prominent by the latter method, as well as by the ferric-chlorid and hematoxylin method. Bacterial stains for tissue sections in which the principles of Gram's stain are used failed to stain the blastomycetes, tho bacteria were well demonstrated. Solutions of thionin (including carbol thionin) and of methylene blue are often useful for the study of the details of the cytoplasm of the organisms, particularly in the study of exudates, etc., in moist or fixed preparations. In sections of fixed tissues, however, they are not satisfactory.

Carbolfuchsin staining, when the stronger acids were used in the decolorization, was productive of nothing worthy of note, all structures giving up the primary stain. When, however, the differentiation was carried out with very weak acids or with plain alcohol, a curious result was observed in the sections of the skin lesion under particular consideration. These were counterstained with alcoholic methylene blue and directly cleared with xylol. Among the great numbers of blue-stained parasites there was seen here and there an individual made very prominent by its intense bright-red color. This was due entirely to retention of the primary stain by the protoplasm, and under no circumstance had the cell-membrane itself shown evidence of acidfastness. These acidfast bodies varied considerably in size and apparent maturity and showed nothing which might suggest an explanation of this peculiar staining reaction.

[13] Jour. Med. Research, 1904, 13, p. 53

[14] Lo Sperimentale, 1912, 66, p. 1.

There may be an analogy between this observation and that of Wolbach[13] that in the coccidioidal granuloma an occasional sphere retains Scharlach R.

The anilin-blue connective-tissue stain of Mallory was the staining principle which, of all those tried, gave the most constant and striking results in the demonstration of the blastomycetes in general. Modified as to length of application, it was the only stain which clearly demonstrated the parasites to be described.

Since this work was done, a study of the organisms from a case of blastomycotic dermatitidis by Bowen and Wolbach[15] has been made accessible. These authors, tho they do not define the exact technic used, also concluded that the connective-tissue stain was the most valuable for the demonstration of the organisms. Hektoen[16] speaks of its use by Wolbach. The only other report in which its use is indicated is that of Rhea, who evidently utilized it in the study of the lesion of a case of fatal blastomycosis reported by Sheperd and Rhea.[17]

Of many variations tried, the technic which proved most satisfactory deviates from that regularly used in connective-tissue-staining in that the primary staining in fuchsin is somewhat decreased, that the counterstain may be not too pronounced, and the secondary staining in anilin blue mixture is greatly prolonged. Tho the usual 20 minutes' staining is sufficient for demonstrating the coarser features of the ordinary capsules, prolonged staining is required that the more delicate capsular structures may be saturated enough to withstand differentiation with 95% and absolute alcohols. When the second stain is applied for but 2 or 3 hours the results are usually less satisfactory than when this solution is used for from 12 to 24 hours. The sections most useful in the study of the forms of the blastomycetes under consideration were treated in this manner. The process stains the outer part of the cell wall a clear brilliant blue, while the protoplasm, appearing yellow to red in color, as do the erythrocytes and tissue cells, shows nothing characteristically differential. As a rule the yeast cells can easily be identified, even with low power lenses, on account of their morphology, their intensity of staining, and the fact that they usually occur within tissue cells or in exudate out of contact with confusing elements.

NOTE ON GENERAL MORPHOLOGY

Study of preparations made by this technic gives one the impression that the usual descriptions of the tissue forms of the ordinary blastomyces are more or less incomplete. In the typical organisms (to be seen in Figs. 6 and 10), one observes immediately surrounding the protoplasm and within the capsule the clear zone which has never been shown to have structure or to retain stains. In moist preparations of blastomycetes this zone seems fluid. The capsule itself, usually described as hyaline and doubly contoured, and not infrequently stated to be homogeneous, is seen to be composed typically of an inner and an outer layer, tho the latter may be absent or indistinguishable. The former, a thin flexible membrane which may be designated the capsula vera, is the true capsule, the essential covering membrane of the

[15] Jour. Med. Research, 1906, 15, p. 167.
[16] Jour. Am. Med. Assn., 1907, 49, p. 1072.
[17] Jour. Cutan. Dis., 1911, 29, p. 588.

organism, on which the outer capsula sclerotica is applied. The latter is often firm and more or less thick and may even be laminated, as in the larger central organism in Fig. 3. In ordinary gemmation the former membrane, which stains a more delicate blue than the capsula sclerotica, does not rupture but evaginates to retain the daughter cell and closes off at the time of separation. The sclerotic layer, on the other hand, often ruptures at the point of budding shortly after the process is inaugurated and remains with the mother cell. At other times, this layer in such instances being rather thin and flexible, both capsules persist about the daughter cell through its formation. This is seen in the largest budding organism in Fig. 5.

It has been interesting to study the process of capsule-formation in sections from subcutaneous lesions of an ordinary case of generalized blastomycosis. In these preparations, specially stained, are seen great numbers of ordinary, thin- and thick-walled, budding and quiescent parasites, together with numerous capsular remnants of dead organisms. These capsules or shells are often seen to persist (Fig. 6, 7, and 8), at times in much the normal form. The protoplasm, however, disintegrates and the shell may be found empty (Fig. 6) or filled with granular debris (Fig. 7). The striking feature of these capsules is the manner in which they increase in size and the extent to which this increase may be carried. It at times appears to begin as delicate knobs or buds of typical blue-staining capsular material within the shell (Fig. 6). Again, it may be less systematically applied, appearing as irregular masses (Fig. 8). In Fig. 7 the body is undergoing an irregular laminar increase. The very large body in Fig. 9 shows both the generally laminated appearance and knoblike protuberances which are themselves laminated. The flower-like body in Fig. 10 is an odd result of the process. In the two endothelial foreign-body giant cells of Fig. 11, are seen masses of the hyaline capsular material undergoing digestion.

Since after the death of a parasite the empty capsule may persist in the tissue or exudate and increase in size, sometimes to a remarkable degree, it would appear to be quite clear, that, as held by certain European writers,[13] the capsula sclerotica is not a product of the cell's vital activity but is a deposit or accumulation of a specialized material applied from without. This possibility has been mentioned by Ricketts,[18] but not emphasized. It is probable that certain of the forms described by Gilchrist and Stokes[1] are of similar nature.

[18] Quoted by Hyde, Jour. Cutan. Dis., 1901, 19, p. 44.

MORPHOLOGY OF ORGANISMS IN THE SPECIAL LESION STUDIED

The unusual lesion found in the skin of the case under consideration, in which were great numbers of organisms in unusual forms, was clearly explained by a study of sections prepared by the prolonged anilin-blue staining. The irregularly coarsely granular appearance of the giant cells and necrotic material was seen to be due largely to the vast number and very small size of the parasites present. The nature and origin of these minute forms were shown by the demonstration about the majority of them of sharply drawn blue-stained capsular membranes. These were often extremely delicate and at times defined only by close scrutiny with the higher-power oil-immersion lenses. Furthermore, it seems probable that not all had membranes sufficiently distinct to resolve. While great delicacy of membrane was the rule, this did not seem necessarily to correspond to the size of the parasite, for some of the smaller sizes, tho not the smallest, possessed relatively thick shells. While the majority of these organisms were rounded, many which presented more irregular outlines were scattered throughout the lesion. The most of these irregular bodies which were not explainable by pressure of surrounding structures were more or less pear-shaped, and upon careful study were seen to be miniature budding cells. All stages of daughter-cell-formation might be found. Particularly noticeable was the fact that this activity was not confined to the cells of any one stage of growth or maturation, but was seen in individuals of all sizes and forms. This may be observed by close scrutiny of Figs. 2 to 5 inclusive. It was not unusual to find two cells, each but 2 or 3 microns in diameter, just about to complete the fission process by separating, each of the daughter cells being practically identical in size and appearance and both covered by the thinest discernible capsule. Many such dividing cellules would have been considered two distinct organisms had it not been for the demonstration of the common cell membrane. Another small, but apparently more mature, type of cell, measuring from but 5 to 10 microns in diameter, might show a very thin-walled bud springing from an aperture in the sclerotic layer of a cell wall as heavy as ordinarily seen on the large sclerotic organisms. The active budding process observed often gave the small thin-walled organisms the appearance of motility.

Granule-formation in the cytoplasm was rather constant, the granules being made very prominent by certain stains. The smallest forms seemed always to contain one such condensation of cytoplasm;

in them this seemed often to constitute the entire protoplasm. In larger organisms there might be several granules present, but no significance could be attached to them. The smaller more compact giant cells often contained few but the smallest forms of parasites. The individuals formed earlier seemed progressively, if slowly, to increase in size, while the total numbers of parasites rapidly increased by the active gemmation described. This resulted in a corresponding increase in size of the giant cells. Ultimately these cells, filled with organisms showing a wide range in size, underwent disintegration. Fig. 2 shows 3 stages of this process.

DISCUSSION

The early and rapid multiplication of the organisms described, by the fission of individuals even of very small size, explains the appearance of great numbers of these spore-like cellules in the lesions in which no endosporulation was found. While budding forms of the ordinary type were numerous, they were, as argued by LeCount and Myers in their case, far too few to account for the vast numbers of minute organisms found. The degree of deviation from the normal process is more clearly appreciated when one compares the size and numbers of the bud processes in the ordinarily active blastomycotic lesion with those in the tissue described.

Study of the lesion in specimens stained by common methods did not clearly eliminate the possibility of the cellules' being formed by endosporulation. Not only were the minute budders unappreciated, but the appearance of clusters of stained granules in old broken capsules, giving the suggestion of sporulation, was somewhat misleading. In the special preparations, however, tho there could be found any stage between the smallest cellule on the one hand, and either a mature thick-walled cell or a large thin-walled organism on the other, nowhere was there found evidence of true endosporulation.

That the organism described may be a new variety of the blastomyces is held improbable. If the forms found in all the lesions were of the peculiar type described, the possibility of its being a new species would be worthy of consideration. There was, however, an apparent localization of the unusual multiplication activity in the skin focus, which is similar to the limitation to the cerebellar focus of the small forms in LeCount and Myer's case. This feature is in accord with the liability to morphologic variation of the blastomyces under different influences, particularly apparent when the organisms are arti-

ficially cultivated. While descriptions of lesser variations are numerous, Stober reports[19] that in a culture contaminated with B. subtilis he observed form variations in an otherwise ordinary strain that seemed even to have attained true sporulation. These and other facts make it probable that the unusual morphology of the organisms in the skin was due to temporary variations under the influence of unusual biologic conditions.

What the factors were which influenced this frantic multiplication cannot be asserted. The blastomycotic nature of the infection was not suspected clinically and was demonstrated only by the study of routine sections some weeks after the autopsy. But one piece of skin had been preserved and this had been removed only because the inguinal lesion did not, as did those elsewhere, resemble entirely that of pellagra. In view of the probable pellagrous condition and of the tuberculous infection of the lung, it seems likely that the blastomycotic invasion was secondary, apparently by way of the skin eroded by the pellagrous dermatitis. It is at least interesting to consider the possibility that this little-understood condition should so markedly modify the activity of the blastomyces. It is also not clear why the distribution of the blastomycotic lesions should not have been more general, in view of the probable ease of dispersion of the small form of organism found.

CONCLUSIONS

The description of the lesion considered resembles closely those of but one or possibly two lesions which have been carefully described in reports of other cases of systemic blastomycosis.

In this lesion were very numerous organisms of varying sizes, occurring diffusely and in clusters within the endothelial and giant cells and in necrotic cell debris, many of which were so small and so massed as to suggest strongly their formation by a process of endosporulation.

When specially stained, the small organisms were shown conclusively to have been formed by the ordinary budding process. This was here extremely active and, contrary to the ordinary condition, was seen in organisms of even the smallest sizes, and apparently of the most recent formation. It is not thought to indicate a variation in the species of the blastomyces.

Tho no explanation can be advanced for the frantic multiplication, it can be suggested as possibly significant that the blastomycotic infec-

[19] Arch. Int. Med., 1914, 13, p. 509.

tion was apparently secondary in skin lesions of a patient clinically a distinct pellagrin, in whom was also found pulmonary tuberculosis. The possibility that the causative factor of the pellagrous dermatitis may have also supplied the unusual stimulating influence to the blastomyces is interesting to consider.

After many attempts to stain these organisms differentially a slight adaptation of Mallory's anilin-blue connective-tissue stain was found to demonstrate them most clearly by delicate blue-stained capsular membranes. This stain, first advocated by Bowen and Wolbach, seems to be the most satisfactory process for the demonstration of the blastomycetes in the tissues. It proves useful in demonstrating details of the organisms ordinarily determined with difficulty, and in locating organisms in certain lesions where they are scarce or partially degenerated. It should be more generally utilized in the study of the lesions of this and similar infections.

For the sake of accuracy of description it seems well to emphasize the typical compound structure of the capsule of the blastomyces in the tissues, consisting as it does of an inner delicate capsula vera and usually an outer applied capsula sclerotica. The extraneous source of this outer capsular material is demonstrated by the progressive increase in size of a capsule after death of the parasite.

EXPLANATION OF PLATES

PLATE 22

FIG. 1. Field from central area of a skin lesion in the case described, showing the numbers, size, and coarsely granular appearance of the endothelial giant cells, and the practical absence of leukocytes of other types. Destruction of the normal subcutaneous tissue is evidenced by poorly supported blood vessels among the cells of the granuloma. Hematoxylin and eosin. Low dry lens.

FIG. 2. Two large giant cells, showing their coarsely granular character, which is due largely to the great numbers of very small and larger parasites, the majority too small and delicate to be clearly defined. An old disintegrating giant cell containing several organisms of larger size is partly shown.

FIG. 3. Field from similar lesion, with organisms apparently not so numerous and averaging considerably larger than in Fig. 2. Several budding forms, with walls varying markedly in thickness, can be seen.

PLATE 23

FIGS. 4 AND 5. Other fields of the same lesion illustrating variation in the size of the organisms. Close scrutiny demonstrates particularly well the gemmation of very small forms and the extreme delicacy of the capsular membranes of the smaller organisms.

Figs. 2 to 5 inclusive were prepared by prolonged staining with Mallory's connective-tissue stain. Bausch and Lomb 1/16 oil immersion lens.

FIGS. 6 TO 11. Nonvital deposition growth of the capsula sclerotica in dead blastomycetes. Fig. 6 compares two normal organisms with large attenuated empty capsules which show four delicate in-growing knobs of capsular material. Fig. 7 shows the early laminar deposit upon a broken capsule filled with granular debris. The blastomycete in Fig. 8 contains small masses of apparently unattached hyaline material among the debris. The body shown in Fig. 9 is very large and shows both laminar and knoblike increase. The odd flower-like body in Fig. 10 is unusual. In Fig. 11 two endothelial giant cells enclose and are digesting such hyaline bodies.

PLATE 22

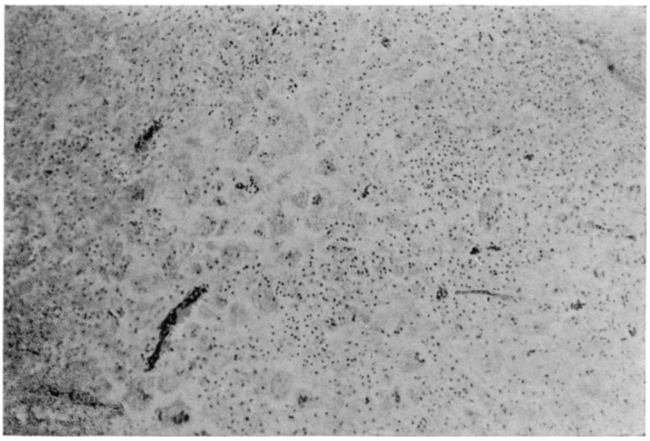

Figure 1

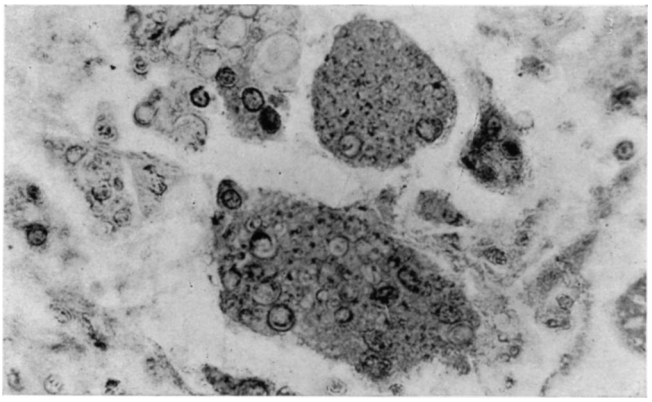

Figure 2

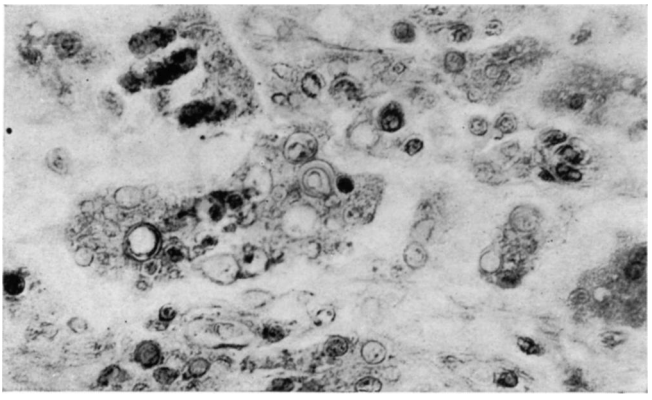

Figure 3

Plate 23

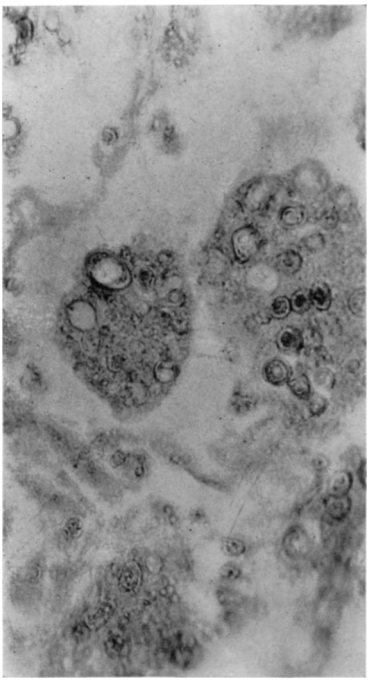

Figure 4

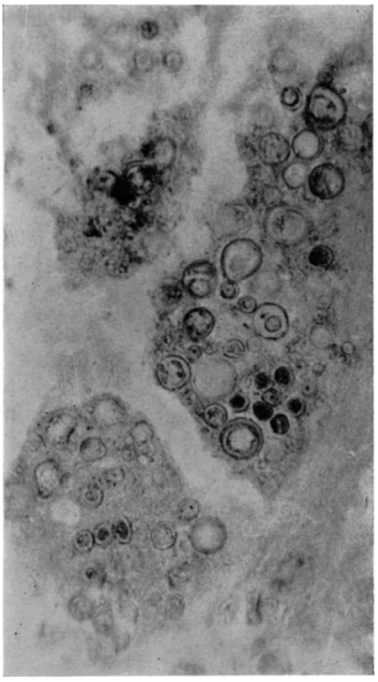

Figure 5

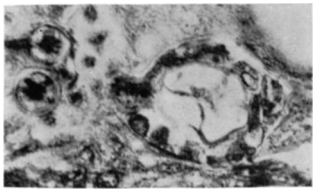

Figure 6

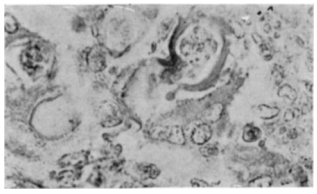

Figure 7

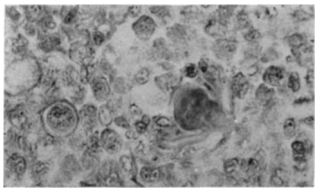

Figure 10

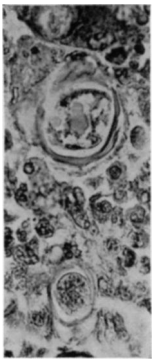

Figure 8

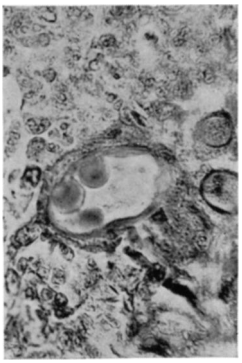

Figure 9

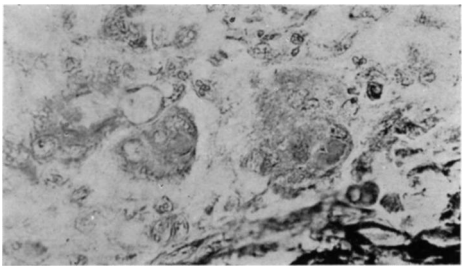

Figure 11